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Prospective assessment of autism traits in children exposed to antiepileptic drugs during pregnancy

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Author contributions

AW, VA, DR, FV contributed to conception and design, CN and SB to acquisition of data, CN and AW to analysis and all authors contributed to interpretation of data, revision and final approval of the manuscript.

Summary

Purpose: The association between autism spectrum disorders (ASD) and prenatal anticonvulsant exposure is increasingly investigated, but comprehensive, blinded assessment using a validated instrument for autism within a well-characterised prospective cohort has not been conducted. Thus, existing studies may represent an underestimate of the true risk. Here we present a prospective cohort study in children exposed to anticonvulsants during pregnancy, with all assessments conducted blind to drug exposure status.

Methods: Participants were 105 Australian children aged 6-8 years who were recruited via the Australian Pregnancy Register for Women on Antiepileptic Medication. Maternal epilepsy, pregnancy and medical history data were obtained prospectively. Autism traits were assessed using the Childhood Autism Rating Scale (CARS).

Key findings: Eleven children (10.5%) had elevated CARS scores. Two were exposed to valproate monotherapy (2/26; 7.7%), two to carbamazepine monotherapy (2/34; 5.9%) and seven to valproate in polytherapy (7/15; 46.7%). Linear regression analysis showed that the mean valproate dose during pregnancy was a significant predictor of CARS scores after controlling for polytherapy, mean carbamazepine dose, folic acid use, seizures during pregnancy, tobacco and marijuana use, maternal IQ and socioeconomic status. First trimester folic acid supplementation and marijuana use were also significant predictors of CARS scores.

Significance: Using direct assessment of children in our prospective study, we found an elevated rate of autism traits across the sample. The most important determinant of

association with autistic traits was higher doses of sodium valproate exposure. The use of valproate in women who may become pregnant is now generally avoided, however there are insufficient data regarding the risk of ASD with low-dose valproate. If this risk is no greater than other AEDs, it may enable women with genetic generalised epilepsy to retain optimal seizure control as well as minimize harm to their unborn child.

Keywords: Prenatal; autism spectrum; valproate; neurodevelopment; anticonvulsant

Introduction

Antiepileptic drugs (AEDs) are a group of commonly prescribed medications used to treat epilepsy, bipolar depression and severe pain. Fetal exposure to AEDs is known to increase the risk of congenital malformations and intellectual impairment[1-3]. Sodium valproate appears to carry a heightened risk, particularly at higher doses[4; 5]. Early concerns about a possible link between fetal exposure to AEDs and neurobehavioural problems, specifically autism spectrum disorders (ASD) arose from case report, retrospective and animal studies[6-10]. More recently, Bromley and colleagues[11] found an increased rate of ASD amongst children exposed prenatally to AEDs, with seven (2.8%) of the 249 AED-exposed children in their sample receiving diagnoses independently of the study[11]. The majority were exposed to valproate monotherapy (6.3% of the group exposed to valproate monotherapy) or valproate in combination with other AEDs. Of note, however, this paper reported cases for whom a diagnosis was made separately to the study, by the child's health-care professional; no formal assessment was undertaken as part of the study protocol. It is therefore likely that the incidence of ASD was underestimated. Nevertheless, their findings were echoed in a data-linkage study[12] which reported an absolute risk of 4.15% for ASD and 2.95% absolute risk for childhood autism in children exposed to sodium valproate compared to all children of women with epilepsy. The Norwegian Mother and Baby Cohort also found an elevated risk of autistic traits in children exposed to AEDs using maternal assessments of the child at three years of age (6.0% across all exposures compared to a reference rate of 1.5%). They found significantly elevated rates in children exposed to monotherapy (5.6%) but not polytherapy, and this reflected a particular increase in children exposed to lamotrigine (9%) but not valproate or carbamazepine[13]. The lack of dose information in the latter

study prevents further interrogation of the pattern of results for valproate, which differs from previous studies.

No study to date has conducted blind assessment of autism traits using a validated instrument in a cohort of children exposed to AEDs in whom prospectively collected data on pregnancy and epilepsy variables are available. A better understanding of the incidence of ASD in children who have had prenatal AED exposure is important for clinical management of women with epilepsy and their families. A lack of systematic assessment or control for confounding variables may lead to either under- or over-estimation of the true incidence of ASD associated with prenatal AED exposure. This in turn has direct implications for the management of women with epilepsy. The aim of the current study was to systematically investigate rates of ASD, using objective assessments in a prospectively-recruited AED-exposed sample. We adopted a pragmatic approach that used a reliable screening tool rather than extended assessment, given our broader study protocol. We predicted that children prenatally exposed to AEDs, particularly valproate, would exhibit elevated rates of ASD in comparison to the general population, which is similar in our Australian setting (0.63%[14]) to global estimates (0.62%)[15].

Methods

Participants: Women with epilepsy and their children were recruited through the Australian Pregnancy Register for Women on Antiepileptic Medication (APR)[4]. The APR commenced prospective data collection about AED use by pregnant women in 1999. The register is voluntary, and women across all Australian states and territories are eligible to participate; all contact is by telephone. A variety of methods are used to inform women

about the register, including by their treating medical practitioners, nurses, or allied health professionals, or by other pregnant women and national advertising (website, lay organisations for epilepsy). Following written informed consent, relevant details are obtained from pregnant women on recruitment (usually in the first or second trimester), at 7 months of pregnancy, in the first postnatal month, and at the end of the first postnatal year. Treating doctors are contacted to confirm medical details. Fetal malformations are classified according to the Birth Defects Registry of Victoria [16]. Children with major birth defects or a diagnosis of epilepsy were excluded, as these conditions are known risk factors for ASD[17; 18].

Between November 2007 and May 2010, 175 women with 190 children aged six to eight years were identified from the APR and invited to participate in the study. Mothers of 27 children (14%) declined to participate. The most common reasons for non-participation were concerns about their child's ability to cope with testing (33%), inability to meet the time commitment (26%), and maternal illness (11%). Mothers of two children were deceased, and two had moved overseas or to inaccessible areas of Australia. Mothers of a further 48 children expressed interest in the study but were not seen because an appointment was unable to be scheduled during the study period. Of the remaining 111 children (including two sets of twins), six were excluded; two had developed epilepsy, two had malformations detected after one year of age, and two were not exposed to AEDs. The remaining 105 children comprised 26 who were exposed in utero to sodium valproate, 34 to carbamazepine, 11 to other AEDs in monotherapy and 34 to AED polytherapy. Demographic details of participating families are provided in Table 1. For women taking polytherapy, the combination of drugs for those taking valproate (N=15) and those who did not (N=19) is shown in Table 2. The mean age of assessment was 7.4 years (SD=0.6 years).

[INSERT TABLE 1 ABOUT HERE]

[INSERT TABLE 2 ABOUT HERE]

There was no significant difference between mothers of the children reported here, and those who did not participate in maternal age, frequency of drug type, or frequency of epilepsy type. The mean gestational age of children born to mothers who did not agree to participate (38.53, SD=2.79 weeks) was significantly lower than those who did participate (39.35; SD=1.78 weeks), and this was due to a significantly higher proportion of preterm births in the non-participatory group (χ^2=8.9, p=.003).

Procedures: Maternal epilepsy, pregnancy and medical history data were obtained from prospectively-collected records. Maternal and paternal demographic data were collected, and maternal IQ was measured with the Wechsler Abbreviated Scale of Intelligence[19]. Family socioeconomic status (SES), as measured by the highest maternal or paternal occupational level, was rated according to the ANU4 Scale[20]. Dichotomous variables were created for maternal polytherapy, folic acid and marijuana use, and the presence or absence of seizures during pregancy. Children participated in a neuropsychological examination, including assessment with the Childhood Autism Rating Scale (CARS)[21]. All assessments were conducted blind to drug exposure status of the child and clinical diagnosis of the mother. Assessments with the Childhood Autism Rating Scale (CARS) were conducted by two authors (CN, SB) and consensus meetings conducted (CN, SB, AW) to confirm scoring.

The CARS is a commonly used clinician-rated behaviour observation scale. Scores range from 15 to 60, with scores of 30 or higher consistent with a diagnosis of autism. Scores above 27 also raise concern; children with Pervasive Development Disorder Not Otherwise Specified (PDD-NOS) have been reported to achieve mean CARS scores of 28.3 ± 4.2[22]. The CARS demonstrates good reliability and a high degree of correlation with DSM-IV diagnoses[21-23]; the relationship with DSM-V has not been reported to date. However, recent research found that CARS diagnoses correspond well with clinician diagnoses and the Autism Diagnostic Observation Schedule (ADOS)[24] and the Autism Diagnostic Instrument-Revised[25] and importantly the CARS was developed to differentiate ASD from other neurodevelopmental conditions, particularly intellectual disability. In the current paper, we use the term 'autistic traits' to refer to any child whose score on the CARS was equal to, or exceeded, 27 points. The ethics review boards of the coordinating institutions approved the study, and informed written consent was obtained from all women before participation.

Statistical Analyses: Group statistics were calculated using Chi-square analysis for frequency data. Analysis of variance was used to compare scores across drug-exposure groups. Due to unequal group sizes, Mann-Whitney U was used to compare relevant scores in those with and without elevated CARS scores. Linear regression was used to examine the contribution of relevant variables (i.e. those identified in univariate analyses or based on findings in previous research) to outcomes of interest.

Role of the funding source

The sponsors of the study had no role in study design, data collection, data analysis, data interpretation, the writing of the report, or in the decision to submit the paper for publication. The corresponding author had full access to all the data in the study and had

final responsibility for the decision to submit for publication.

Results

Of the 105 children exposed to AEDs in utero, 11 children met the criteria for autistic traits. Of these, eight children obtained scores higher than 30 on the CARS (placing them in the ‘autistic’ range), and another three displayed CARS scores over 27 (i.e. on the ‘concern for autism’ range). Four of these children (3.8% of the total sample) had a previous diagnosis of ASD. One further child, exposed to valproate monotherapy, came to the study with an existing diagnosis of autistic traits, yet his high level of intellectual and language abilities placed him below the CARS cutoff, and he is not included in the analyses presented here. Two of the 11 affected children (7 boys, 4 girls) were exposed to valproate monotherapy, two to carbamazepine monotherapy and seven to valproate in combination with one or more other drugs. Details are provided in Tables 3 and 4.

[INSERT TABLE 3 ABOUT HERE]

Group comparisons showed that, compared to mothers of children without autistic traits, mothers of children with autistic traits were more likely to have had one or more seizures during pregnancy (p=.010, Fisher’s exact test), more likely to have used marijuana during pregnancy (p=.028, Fisher’s exact test), and less likely to have taken folic acid supplements in the first trimester (p=.028, Fisher’s exact test). Maternal IQ and family SES were also lower in the autistic traits group, but these differences were not statistically significant (Maternal IQ U=670.5, p=0.11; SES U=653.5, p=0.084). Nevertheless, given the known association in the general population between these

variables and child IQ[26 27] and the importance of these variables for long-term outcomes in children born to women with epilepsy[3], these variables were retained for regression analyses. No group differences were found with regards to alcohol, tobacco, caffeine, or other drug use during pregnancy, preconception folic acid, parental age, or paternal education, epilepsy type, length of breastfeeding, pregnancy complications, birth type, anaesthetic use, or number of children born prematurely.

[INSERT TABLE 4 ABOUT HERE]

CARS scores of children exposed to valproate (N = 26), carbamazepine (N = 34), lamotrigine (N = 9), polytherapy with valproate (N = 15) or polytherapy without valproate (N = 19) were compared. Specific details of the combination of medications used by women in the latter two groups are shown in Table 2. Children exposed to polytherapy with valproate scored significantly higher than all other groups, (mean difference = 7.55-9.42, $p<.001$). No other group differences were significant. Mean scores for each group are shown in Figure 1.

Repeated measures analysis of variance showed that the dose of valproate did not change significantly across pregnancy in the monotherapy (using the Huynh-Feldt correction for non-sphericity; $F(1.6, 40.4)=3.14$, $p > 0.05$) or polytherapy groups (corrected $F(1.1,16.1)= 1.33$, $p>0.05$). Thus, mean dose of valproate across pregnancy was used to examine the relationship with CARS scores across all participants exposed to valproate. This was, however, not significant ($r=0.30$, $p=0.06$, two-tailed). Mean dose of valproate was significantly higher in polytherapy (Mean = 1589, SD=986.66) versus monotherapy (Mean=961.78, SD=629.74) exposures ($t(20.7)=-2.22$, $p = 0.04$). Despite

this, there was no relationship between CARS scores and VPA dose within the valproate polytherapy group (r=-.06, p> 0.1), but the association between dose and CARS scores was significant in those exposed only to valproate (r=0.61, p=0.001). There was no significant difference between the overall mean carbamazepine dose in the monotherapy (Mean=723.02, SD=447.86) and 'other' polytherapy groups (Mean=917.15, SD=315.98), t(46)=1.47, p > 0.05.

Carbamazepine doses did increase significantly across pregnancy within the monotherapy group (corrected F(1.5,48.5)=5.00, p=0.018). Nevertheless, CARS scores were not significantly related to dose in the first, second or third trimester (all p > 0.1).

[INSERT FIGURE 1 ABOUT HERE]

Linear regression analysis (Table 5; adjusted R^2 = 0.42, F (8, 92) = 9.95, p < .001) showed that mean valproate dose significantly predicted CARS scores after controlling for polytherapy, mean carbamazepine dose, folic acid use, tobacco and marijuana use, seizures during pregnancy, maternal IQ and SES. Use of folic acid in the first trimester and marijuana use were also significant predictors of CARS scores. The overall model and specific predictors remained unchanged when either of the siblings with elevated CARS scores was removed from the analysis. The regression analysis was repeated with valproate and carbamazepine exposure included as binary variables (i.e. not dose variables), and the overall model and the specific predictors remained significant.

Discussion

Here we report the first direct, prospective evaluation of autistic traits in a cohort of children exposed to AEDs in utero. In addition to children who had an existing ASD diagnosis when they entered the study, we identified another group whose CARS scores show they have autistic traits that warrant clinical investigation and support, demonstrating the importance of long-term follow up in children whose mothers required AEDs during pregnancy. The overall proportion of affected children in our sample is substantially higher than the estimated prevalence of ASD in six to twelve year-olds nationally or internationally. Children who had in-utero exposure to valproate were most likely to have elevated CARS scores, with 7.7% of the valproate monotherapy group and 46.7% of the valproate polytherapy group displaying ASD symptoms. The dose of valproate taken during pregnancy was found to be an independent risk factor for elevated CARS scores, while polytherapy *per se* was not. CARS scores were not elevated in children exposed to polytherapy without valproate suggesting that valproate, or valproate dose, rather than polytherapy *per se* is the critical determinant of the relationship, an observation that requires verification in future studies. The observation of a dose-response relationship within those exposed to valproate in monotherapy suggests a role for valproate in ASD risk. This finding is in line with studies investigating the risk of other adverse outcomes (i.e. birth defects and neurocognitive deficits) in children who have had prenatal valproate exposure, which have also been found to have a strong relationship to dose[1; 5; 28-32], with higher dose associated with poorer outcomes than low dose. There is not, however, a single agreed boundary at which 'low' versus 'high' is drawn. Recent data from the UK long-term follow-up study highlighted concern about verbal abilities and educational needs in children exposed to low-dose valproate [33], which could suggest that there is no 'safe' dose for valproate. This group is an important one, clinically, and

additional neurodevelopmental research is required to clarify the consequences of low-dose valproate exposure on the unborn child.

There is not currently strong evidence for neurodevelopmental risk associated with intrauterine exposure carbamazepine. Recent data found subtle risks associated with this AED[34], however the Cochrane consortium suggested recently that any association between poor outcome in younger children exposed to carbamazepine was lost when variability in studies was taken into consideration[3]. In our cohort, the proportion of children exposed to carbamazepine monotherapy with elevated scores (5.9%) was higher than the general population, and at a similar level to valproate monotherapy. Nevertheless, children exposed to polytherapy without valproate were most often exposed to carbamazepine and yet did not show elevated rates of autistic traits, and carbamazepine dose was unrelated to CARS scores. Thus, our data should be interpreted with caution and additional studies are required before changes in practice with regards carbamazepine in pregnant women should be considered.

Autism spectrum disorders are highly heritable, with increased risk in first-degree relatives. People with ASD may also have seizures and there are epilepsy syndromes in which ASD is a prominent feature, yet the likelihood of inheriting ASD from a parent with epilepsy above and beyond population rates of inheritance is relatively less well understood. At interview, no mothers of children with elevated CARS scores reported a maternal or paternal family history of autism, although we did not screen parents to verify this. Some children with elevated CARS scores had affected siblings who were not seen as part of the study, and this might be interpreted as familial risk. In all cases, however,

mothers reported that the siblings were also exposed *in utero*, which suggests that AED exposure rather than 'genetic susceptibility' explains the occurrence of autism traits in those families. The different rates of elevated CARS scores across the AED exposure groups in our study suggest that maternal epilepsy *per* se does not account for autism traits in their offspring, and recent data from women with epilepsy who did not take AEDs during pregnancy found no evidence for elevated autism risk[13]. Genetic factors may *mediate* the association between valproate exposure and poor neurodevelopmental outcome and this may be dose-dependent and additional studies are required to clarify this.

A number of other factors may influence the likelihood of a child meeting the criteria for ASD following in utero AED exposure. Some have speculated that increased use of maternal folate may be associated with rising ASD prevalence[35]. Our data, however, contradict this and in fact highlight the importance of folic acid supplementation in women with epilepsy, with an association between elevated CARS scores in children whose mothers did not report folic acid supplementation in the first trimester. We also found a clear association of maternal marijuana use during pregnancy and ASD symptoms in the children. These two factors should be included in preconceptual counselling and ante-natal care discussions with women with epilepsy.

The results of this study extend previous reports of increased rates of ASD following fetal valproate exposure[7-9; 11; 12] by providing the first prospective assessment of all children in a cohort exposed to anticonvulsant medications *in utero* and controlling for confounding variables. These data are consistent with studies showing that autism-like behaviours can be seen in the offspring of pregnant rodents administered valproate[10]. Our findings

support the concluding remarks of Bromley and colleagues[11], that their study methods may have underestimated the risk of ASD risk in AED-exposed children. In addition to the younger age of their sample, almost a third of whom were below six years old, their study did not systematically assess ASD with an instrument such as the CARS, reducing the detection rates. The prevalence of autism traits in the Australian population when using the CARS is not known, so one interpretation of our findings is that our data simply reflect a greater sensitivity to ASD but poor specificity or poor diagnostic accuracy as a result of using this instrument in all cases. A recent systematic review of diagnostic and screening instruments for autism noted that any instrument with a diagnostic accuracy of 80 percent or more is equivalent to the "gold standard" of multi-disciplinary team diagnoses[36]; the CARS exceeds this threshold and has excellent levels of sensitivity and specificity. Thus, the current data raise the concern that many children exposed to AEDs in utero have undetected neurodevelopmental problems. Importantly, our sample differs from previous studies[7-9; 11] in that we excluded children with major birth defects. There is strong evidence that fetal valproate exposure is associated with elevated rates of birth defects[37], and that birth defects and autism often co-occur[17]. Our data suggest that there is an elevated risk of ASD independent of the presence of major birth defects and highlight the importance of continued monitoring of exposed children beyond infancy, particularly in those for whom initial surveillance is unremarkable.

Sample size and selection bias are important factors to consider in observational cohort studies such as ours. The size of our individual groups was relatively small which may result in a misrepresentation of the risks associated with specific prenatal AED exposures. This would most likely occur if there was selection bias in our study. One potential source of bias in this study is the possibility that women who held concerns

about their offspring were more likely to agree to participate. The study cut-off date precluded assessment in sixty-one percent of those approached about participation but not included in the study. Of the remaining group (14%) who declined to participate, the most common reason given was maternal concern about the child's ability to cope with testing (anecdotally due to perceived educational or behavioural difficulties in the child). Mothers who did not agree to participate were similar to those included here on key epilepsy variables. The only characteristic that differed between participators and non-participators was gestational age, with more preterm children amongst those who *did not* take part. Thus despite our relatively small overall and sub-groups' sample size and the sampling method, the possibility of selection bias in favour of ASD seems unlikely to account for the results we report here. The number of children with elevated scores was relatively small compared to the overall sample. This might impact on analysis of confounding variables, although non-parametric analysis was used to mitigate against this risk.

Another important potential confound is the impact of including in our analyses children with intellectual disability. The CARS was developed to distinguish between autism and neurodevelopmental delay due to other causes and as such performs well in low-functioning children. Items rely most heavily upon the social and behavioural features of ASD and although some of our children performed poorly on IQ tests, their clinical presentation, captured by supra-threshold CARS scores, supports the contention that a higher-rate of ASD is associated with AED, particularly VPA, exposure *in utero*. Impaired social communication skills were often observed in the children in our study who did not already have a diagnosis of autism, suggesting that there is a broad behavioural phenotype that extends beyond cognitive impairment. Our data analyses excluded one

child who enrolled in our outcomes study that had a previous diagnosis of 'autistic traits', yet his high level of function placed him well below the CARS cut-off, including that published recently for use in high-functioning ASD[38]. Including that child in our analyses would show that 11.5% (3/26) of VPA monotherapy exposed children had ASD. Accordingly, the current rate of ASD associated with prenatal AED exposure remains an estimate only; our study highlights the risk profile of this group of children and points to a need for ongoing prospective research in large samples of exposed children.

There is considerable evidence emerging from these and other data that fetal valproate exposure is associated with increased risk of ASD. This under-recognised outcome has significant implications for clinical management of affected women. Women taking higher doses of valproate, particularly in polytherapy, have a greater risk of having an affected child. Valproate remains a highly efficacious drug for patients with primary generalised epilepsy and any decision to change or cease medication must be carefully weighed against the risks posed by inadequate seizure control to the mother or to the unborn child. In women of child-bearing age with epilepsy for whom the risk-benefit ratio is felt to favour continuing valproate treatment during pregnancy, the dose should be kept as low as possible to minimise the risk of the child having ASD in view of the strong associate between valproate dose and CARS found in this study. Many patients with genetic generalised epilepsy can be controlled with low dose valproate (<1000 mg/day), and in some this is the only treatment that can control the seizures. While low dose valproate treatment in pregnant women is free of the increased risk of ASD requires further investigation.

Additional study of the link between ASD and fetal AED exposure is required to elucidate underlying mechanisms. In keeping with the elevated risk profile for children of women taking AEDs during pregnancy, especially those from lower socioeconomic groups, there is a need for research that focuses on early detection and early intervention for affected families. The potential for modifications of risk factors in women with epilepsy who become pregnant, including treatment modifications and lifestyle changes, to reduce the risk of their children having ASD[39].

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Declarations

Dr Wood confirms that she had full access to all the data in the study and had final responsibility for the decision to submit for publication.

We confirm that we have read the Journal's position on issues involved in ethical publication and affirm that this report is consistent with those guidelines.

Author TOB has received support for the conduct of work for the APR from Sanofi-Synthelabo; UCB Pharma; Janssen Cilag; Novartis; Pfizer; Glaxo; RMH Neuroscience Foundation and has served as a paid consultant Scientific Advisory Board, Janssen Cilag; speakers fees UCB Pharma, Janssen Cilag, Sanofi-Synthelabo;

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Author DR has served as a paid consultant for Speakers fees, UCB Pharma.

Author AW has not received support directly but declares that she is a member of the Australian Pregnancy Register Executive, which receives support from pharmaceutical companies (Sanofi-Synthelabo; UCB Pharma; Janssen Cilag; Novartis; Pfizer; Glaxo) and RMH Neuroscience Foundation.

All other authors declare no conflict of interest.

We confirm that we have read the Journal's position on issues involved in ethical publication and affirm that this report is consistent with those guidelines

Key Points:

- Prospective, blinded assessment of autism in children exposed prenatally to antiepileptic drugs reveals increased risk relative to population rates
- Children of mothers who took valproate in combination with other antiepileptic drugs are at greatest risk of autism
- There is a dose-response relationship between valproate monotherapy and autism risk
- Additional data on the neurodevelopmental risk associated with low-dose prenatal valproate exposure are required.

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Table Headings:

Figure Headings:

Figure 1

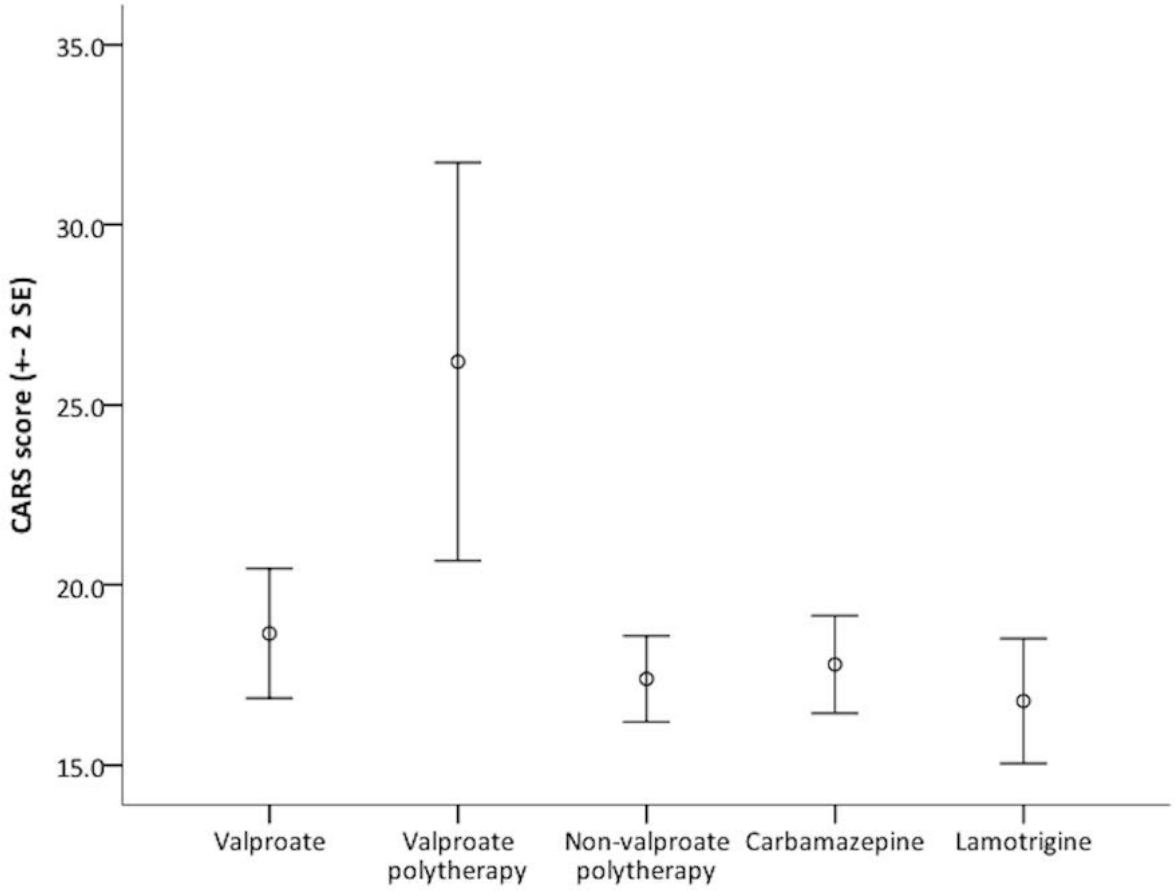
CARS score (+- 2 SE)
35.0
30.0
25.0
20.0
15.0
Valproate
Valproate polytherapy
Non-valproate polytherapy
Carbamazepine
Lamotrigine

Table 1: Maternal characteristics by drug exposure group

	Maternal IQ (Mean(SD)) (range)	**Age at seizure onset (years)**	**Epilepsy onset type (Generalised/ Partial)**	**Seizures during pregnancy (No/Yes)**	**Pregnancy duration (weeks)**	**Child IQ**
VPA (Mono) (N=26)	105.9 (12.8) 83-129	15.6 (6.6) 5-31	21G/5P	20N/6Y	39.4 (1.8) 34-42	95.8 (13.3) 66-117
VPA (Poly) (N=15)	91.7 (14.8) 65-120	11.4 (7.5) 1-28	11G/4P	3N/12Y	39.3 (3.1) 29-41	81.0 (17.5) 40-103
CBZ (Mono) (N=34)	108.7 (12.3) 81-129	16.8(10.1) 0-32	7G/23P/4UK	22N/12Y	39.3 (1.4) 36-41	100.7 (14.3) 71-126
'Other' Non-VPA poly (N=19)	102.3 (14.3) 72-123	12.9 (7.4) 0.5-28	5G/13P/1UK	9N/10Y	39.4 (1.0) 38-41	93.8 (10.6) 72-110

LTG mono (N=9)	109.3 (8.1) 99-119	23.3 (10.3) 1-36	2G/7P	4N/5Y	39.8 (1.7) 38-42	105.1 (5.7) 95-115
Other mono* (N=2)	109.5 (38.9) 109-110	16.0 (8.5) 10-22	2P	1N/1Y	38.5 (0.7) 38-39	96.0 (32.5) 73-119

* One each topiramate, gabapentin. Key: VPA=valproate; LTG=lamotrigine; CBZ=carbamazepine

Table 2: Polytherapy drug combinations in the sample

Polytherapy combinations	Frequency	Percentage
Including Valproate		
VPA + CBZ	1	6.7
VPA + CBZ + CLON	1	6.7
VPA + CBZ + LTG + CLON	1	6.7
VPA + CLON	2	13.3
VPA + LEV	1	6.7
VPA + LTG	6	40
VPA + LTG + CLON	1	6.7
VPA + LTG + ETH	1	6.7
VPA + TIAG	1	6.7
Total	(15)	(100)
Without valproate		
CBZ + CLON	3	15.8
CBZ + CLON + VIG + GAB	1	5.3
CBZ + LTG	5	26.3
CBZ + PHEN	3	15.8
CBZ + VIG	1	5.3
CLON + CBZ	1	5.3
CLON + PHEN	1	5.3
LTG + PHEN	2	10.5
LTG + PHEN + VIG	1	5.3
LTG + TOP	1	5.3
Total	(19)	(100)

Key: VPA=valproate; CBZ=carbamazepine; CLON=clonazepam; LTG=lamotrigine; LEV=levetiracetam; ETH=ethosuximide; TIAG=tiagabine; VIG=vigabatrin; PHEN=phenytoin; TOP=topiramate

Table 3. Rates of elevated CARS scores

	CARS>30	**CARS 27-29**	**Total**
Valproate (Monotherapy)	1/26 3.8%	1/26 3.8%	2/26 7.7%
Valproate (Polytherapy)	6/15 40.0%	1/15 6.7%	7/15 46.7%
Carbamazepine (Monotherapy)	1/34 2.9%	1/34 2.9%	2/34 5.9%
Other (Mono / Polytherapy)	0/30 0.0%	0/30 0.0%	0/30 0.0%
Total	8/105 7.6%	3/105 2.9%	11/105 10.5%

Table 4. Maternal Pregnancy History for Children with Autism Spectrum Disorders or Autistic traits

Case	ASD Details	Child IQ	Antiepileptic Drug(s)	Daily Dose (range across pregnancy)[1]	Other Drug(s)	Folic Acid [2]	Family History of ASD
1	Autism[3] (CARS=35)	66	Valproate	3000mg	Tobacco (25 cigarettes/day) Marijuana (3 joints/week) Alcohol (2-5 units/day until 12 weeks)	500mcg daily AC	No
2	Autism[3] (CARS=32.5)	78	Valproate Levetiracetam	1200mg 2000mg	Tobacco (>30 cigarettes/day) Alcohol (<1 unit/day)	400mcg daily BC	No
3	Autism[3] (CARS=33)	82	Carbamazepine	600-800mg	Paroxetine (dose not specified; until 12 weeks)	None	No
4	PDD-NOS[3] (CARS=38.5)	61	Valproate Clonazepam	2500mg 0.5mg	None	10mg daily BC	No
5	CARS=50;	40	Clonazepam	0.5mg (until	Tobacco (<10	5mg	No

	Intellectual & language delay; Socially unresponsive.		Valproate	21 weeks) 200-400mg (from 21 weeks)	cigarettes/day) Marijuana (3 joints/day until 12 weeks)	daily BC	
6[4]	CARS=37; Intellectual & language delay; Social difficulties.	80	Valproate Lamotrigine	3000mg 100mg	None	None	Sibling (Case 7)
7[4]	CARS=30; Intellectual & language delay; Social difficulties.	64	Valproate Lamotrigine Ethosuximide	2500- 3000mg 50-200mg 500mg (until 4 weeks)	None	5mg daily BC	Sibling (Case 6)
8	CARS=34.5; Intellectual & language delay; Social difficulties.	67	Valproate Lamotrigine	1000mg 400mg (until 12 weeks)	Alcohol (<1 unit/week)	5mg daily BC	Sibling with similar problems[4]
9	CARS=29; Language delay; Social difficulties.	92	Lamotrigine Valproate	400mg 300mg (from 29 weeks)	None	5mg daily BC	Sibling with autism[5]
10	CARS=28; Language	75	Valproate	1000-1500mg	Diclofenac potassium	500mcg	No

	delay; Social difficulties.				(irregular from 28 weeks)	daily AC	
11	CARS=27.5; ADHD[1]; Learning & social difficulties	96	Carbamazepine	1200mg	None	5mg daily BC	Sibling with Asperger's[5]

[1] Where one value is given, the dose was unchanged across pregnancy

[2] AC=Commenced after conception; BC = Commenced before conception

[3] Previously diagnosed

[4] Cases 6 and 7 were siblings

[5] By parental report; child was not seen because they were outside the study age range. All three siblings were also AED-exposed

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Table 5. Predictors of CARS Scores in linear regression

Variable	**B Coefficient (Std. Error)**	**t (p)**
Mean valproate dose	0.002 (0.001)	3.20 (.002)
Folic Acid 1st Trimester	-8.631 (2.830)	-3.05 (.003)
Marijuana Use	14.844 (2.88)	5.16 (<.001)
Mean carbamazepine dose	0.000 (0.001)	-0.07 (.945)
Polytherapy	1.727 (1.126)	1.53 (.128)
Seizure(s) During Pregnancy	0.963 (1.013)	0.95 (.345)
Maternal IQ	0.000 (.044)	0.007 (.994)
Socioeconomic status	-0.014 (0.027)	-0.52 (.607)
Constant	25.831 (5.007)	5.16 (<.001)